

UNC112: A New Regulator of Cell–Extracellular Matrix Adhesions?

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Genetic analyses of *Drosophila* and *Caenorhabditis elegans* have yielded invaluable insights into many basic biological processes, perhaps most notably in the fields of receptor tyrosine kinase signaling and apoptosis. In this issue of *The Journal of Cell Biology*, the identification of UNC-112 as a new cytoskeletal component that may function in the assembly of cell–extracellular matrix adhesions in the muscle of *C. elegans* is described (Rogalski et al., 2000). Since the components of dense bodies of the *C. elegans* muscle are similar to the components of focal adhesions in non-muscle cells, this finding may have broader implications for the assembly of cell–extracellular matrix adhesions.

In non-muscle cells, the assembly of focal adhesions has been extensively analyzed using mammalian fibroblasts. The integrins, which are transmembrane heterodimers, are major components of focal adhesions (Burrige and Chrzanowska-Wodnicka, 1996; Jockusch et al., 1995). In addition, cytoskeletal proteins such as vinculin and talin are localized at these sites. The integrins must engage their extracellular matrix ligands to form focal adhesions. In addition, the cytoplasmic domains of the integrins are important for focal adhesion localization, the tail of the β subunit promoting localization and the tail of the α subunit regulating focal adhesion localization (Burrige and Chrzanowska-Wodnicka, 1996). Cytoplasmic signals, most importantly Rho-mediated contractility, are required for the assembly of focal adhesions in fibroblasts (Schoenwaelder and Burrige, 1999). Finally, one very interesting property of integrins that may be relevant for the formation of these structures is inside-out signaling, a process where cytoplasmic signals act to modulate the affinity of the integrins for their extracellular ligands (Hughes and Pfaff, 1998).

In *C. elegans*, genetic and cell biological approaches have been applied to study the assembly of cell–extracellular matrix adhesions in the muscle of the embryo. Following gastrulation, the embryo elongates to form a worm before hatching. Due to the space constraint of the eggshell, the embryo folds back upon itself twice during the elongation process to form the two- and threefold stages of embryogenesis. Embryonic movement initiates before the twofold stage. Body movements of both the embryo and adult are controlled by four strips of striated muscle, each of which are two cells wide and one cell thick, that lie im-

mediately beneath the hypodermis of the animal. Embryonic movement and elongation were used as phenotypic markers for a genetic screen to isolate mutants defective in muscle development (Williams and Waterston, 1994). A series of *pat* mutants (paralyzed, arrested elongation at twofold) were isolated and mutants from other genetic screens, including *unc-112*, were further characterized and shown to exhibit the *pat* phenotype.

The formation of cell–extracellular matrix adhesions is essential for embryonic movement and elongation, since three mutants exhibiting the *pat* phenotype have mutations in the genes encoding the α (*pat-2*) and β (*pat-3*) integrin subunits and vinculin (*deb-1*). PAT-2, PAT-3, and DEB-1 are highly expressed in muscle cells and are assembled into highly ordered structures (Hresko et al., 1994). Initially, these proteins become localized to the surfaces of the muscle cells that contact other muscle cells and the hypodermis. The PAT-2 and PAT-3 proteins later coalesce to form dense bodies and M-lines, which anchor the actin and myosin filaments of the muscle. DEB-1 localizes to dense bodies, but not to M-lines. UNC-52 (perlecan), a major basement membrane proteoglycan, also localizes underneath the dense bodies and M-lines and is required for the correct assembly of PAT-2 and PAT-3 into these structures. Interestingly, the *pat-10* mutation was found to reside in the *C. elegans* troponin C gene (Terami et al., 1999). Although this mutant exhibits a *pat* phenotype, the myosin and actin components of the muscle appear highly organized (Williams and Waterston, 1994), suggesting that troponin C–regulated contractility is not necessary to promote the assembly of adhesion structures in the muscle.

Rogalski et al. (2000) now report the isolation of the *UNC-112* gene and demonstrate that UNC-112 is expressed in muscle cells of the *C. elegans* embryo, specifically colocalizing with integrins in dense bodies and M-lines. A genetic analysis revealed that PAT-3 and UNC-52, but not DEB-1, were required for the correct localization of UNC-112. The most intriguing finding was that UNC-112 was required for the correct assembly of adhesive structures. Although correctly localized to the muscle cell surface adjacent to the hypodermis during development, PAT-3 failed to form the highly ordered dense body and M-line structures in *unc-112* mutants.

The sequence of UNC-112 reveals extensive homology to the product of the human *mig-2* gene (mitogen-induced gene-2; Wick et al., 1994). This is not to be confused with the *mig-2* gene (cell migration abnormal) of *C. elegans*, which in fact encodes a member of the Rho family of GTP-binding proteins (Zipkin et al., 1997). The human

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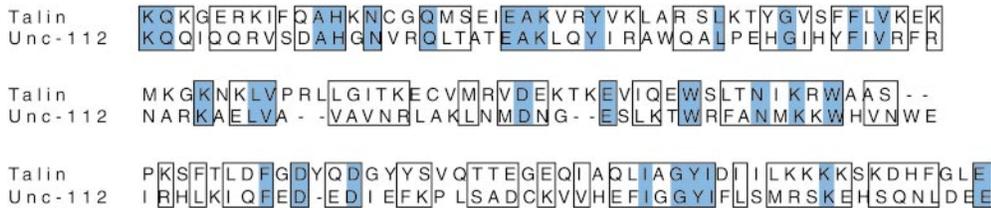


Figure 1. Residues 272 through 412 of talin are aligned with amino acids 573 through 710 of UNC-112. Identical residues are shaded and similar residues are boxed.

mig-2 transcript is induced upon serum stimulation of quiescent WI38 cells, but is otherwise uncharacterized (Wick et al., 1994).

UNC-112 contains a region of homology with the FERM domains of talin and the ERM (ezrin-radixin-moesin) family of proteins (Chishti et al., 1998). Further comparison of UNC-112 and talin using the FASTA sequence analysis program revealed a second region of homology between part of the FERM domain of talin and the COOH terminus of UNC-112 (Pearson and Lipman, 1988; Fig. 1). FERM domains contain binding sites for the cytoplasmic tails of transmembrane proteins and talin and the ERM proteins contain multiple actin-binding sites. Via these interactions, talin and the ERM proteins function to tether the actin cytoskeleton to the membrane (Critchley, 2000; Tsukita and Yonemura, 1999; Mangeat et al., 1999; Bretscher, 1999). In addition to their architectural roles, talin and the ERM proteins may play a role in transmitting cytoplasmic signals since they contain docking sites for signaling molecules.

How does UNC-112 induce the assembly of dense bodies and M-lines? Given the homology to the FERM domains of talin and the ERM proteins, UNC-112 might promote cross-linking of the cytoskeleton to transmembrane receptors. UNC-112 might bind to the cytoplasmic tails of integrins, or alternatively to an unidentified transmembrane protein, and promote cross-linking of the actin cytoskeleton and the formation of dense bodies (Fig. 2). In contrast to this structural role in ordering cell-extracellu-

lar matrix adhesions, UNC-112 may regulate transmission of a signal, targeting either the cytoskeleton or the integrins as part of an inside-out signaling pathway, driving dense body formation (Fig. 2). Although the models presented in Fig. 2 depict the assembly of dense bodies, analogous scenarios can be envisioned for the formation of M-lines. To test these and other possible hypotheses, a number of lines of future investigation are warranted. Reverse genetic approaches will allow an assessment of the role of the talin-homologous sequences of UNC-112 in regulating the assembly of dense bodies and M-lines. Identification of the binding partners of UNC-112 will be an important step in elucidating its mechanism of action and an important area of future investigation will be the regulation of interactions between UNC-112 and its binding partners. Finally, the discovery that UNC-112 plays a role in the assembly of integrin-containing adhesion structures in *C. elegans* will stimulate interest in *mig-2* and other UNC-112-related proteins in mammalian cells. Analysis of these proteins may yield new insights into the regulation of cell adhesion and other integrin-controlled biological processes, such as motility and cell survival. If so, the findings of Rogalski et al. will provide yet another example where genetic analysis of simple model organisms has provided the first glimpse at the function of a protein in mammalian cells.

Research in the author's laboratory is sponsored by grants from the National Institutes of Health (GM53666 and GM5794-3) and the American Cancer Society (RPG-96-021-04-CSM).

Submitted: 14 June 2000

Accepted: 16 June 2000

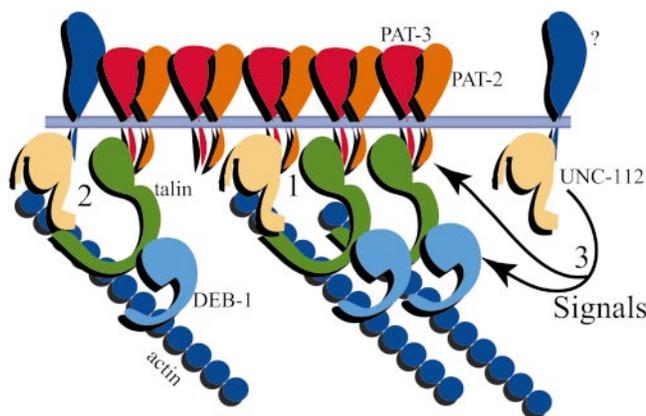


Figure 2. Models of UNC-112 function in dense bodies. UNC-112 might bind integrins (as in 1) or other transmembrane proteins (as in 2) to promote cross-linking of the cytoskeleton and assembly of dense bodies. UNC-112 might stimulate cytoplasmic signals (as in 3) that converge on the cytoskeleton or the integrins to cause assembly of dense bodies.

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